

Electrical Effects accompanying the Decomposition of Organic Compounds.

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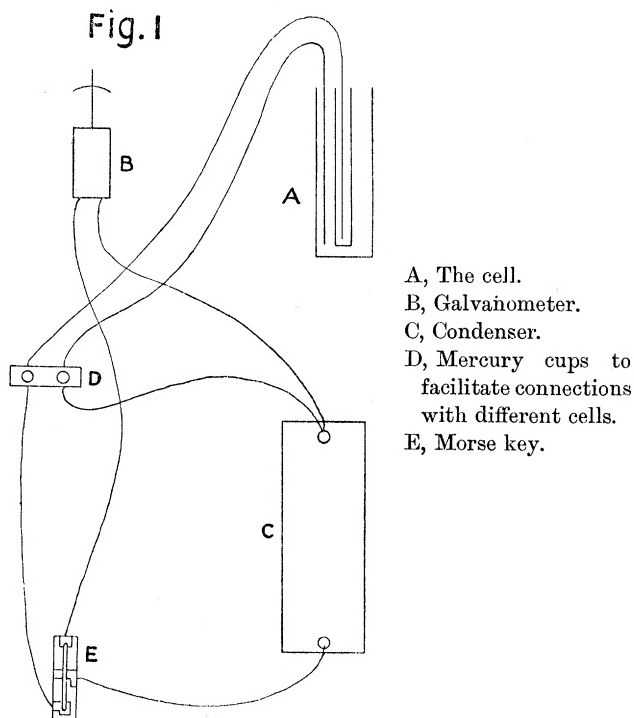
The results of recent researches in electro-physiology have familiarised us with the view that any physiological process accompanied by chemical changes involves an associated electrical change. Haacke and Klein have shown that electrical currents in plants are essentially a manifestation of vital phenomena, and that differences in electric potential are connected both with respiration and carbon assimilation. Waller's investigations have also shown that the excitation of living vegetable protoplasm gives electrical response no less than that of animal protoplasm. He has demonstrated that leaves in a condition of active metabolism give an instant electrical response to the influence of sunlight, which was modified under conditions affecting protoplasmic activity. Apparently almost immediately upon the perception of the stimulus of light, electrical energy begins to be absorbed in the process of photosynthesis. Waller approaches very suggestively the existence of two opposing forces in the presence of analytic and synthetic processes, and recognises that the functions of assimilation and respiration might be mutually antagonistic as regards visible electric effects. His conception that "the product of dissociation . . . gives current *from* the focus of dissociation, whereas a product of association, during its formation, gives rise to a current in the opposite direction," is of great interest.

The line of enquiry now followed lies in the direction only of dissociation, and is a study of electrical effects accompanying fermentation or putrefaction under the influence of micro-organisms such as *Saccharomyces* or bacteria. The special physiological character of fungi or bacteria demands the disintegration of organic compounds as a necessary source of energy, and where there has been absorption of energy in a synthetic process one must look for its liberation when the change is of an analytic nature. The evolution of caloric energy during fermentation or putrefaction is commonly recognised, and that electrical energy is also liberated during these processes is a conception of considerable interest. In this preliminary communication some experiments are described which were undertaken to determine whether any E.M.F. is developed when organic compounds are broken down through the fermentative activity of yeast and other organisms. Cultures of

Saccharomyces cerevisiae and certain species of bacteria were grown in nutrient media, and the chemical action of their vital processes was utilised to develop electrical energy in a manner parallel to the production of E.M.F. by means of the ordinary galvanic cell.

The Apparatus.

The apparatus employed (fig. 1) consisted of a glass jar containing a porous cylinder. The same nutrient fluid was placed in the glass jar and in the porous cylinder, and two platinum electrodes were introduced, one into the



fluid in the jar and the other into the porous cylinder. In the case of pure cultures, instead of the jar, a large boiling tube was used, which could be plugged with cotton wool and the whole apparatus sterilised either by intermittent boiling or in an autoclave. After sterilisation the fluid in the boiling tube could readily be inoculated with the micro-organisms under investigation. Cultures of micro-organisms, when introduced into either the fluid in the jar (*the outer fluid*) or that in the porous cylinder (*the inner fluid*), under suitable conditions, set up a chemical action, and the apparatus so far described constitutes a type of galvanic cell.

Throughout the main experiments platinum was used as the electrode, and

to avoid the necessity of long platinum wires a short length of this metal was soldered with silver to a copper wire. For the purpose of insulation the wire was enclosed in a glass tube, the extremities of which were then fused round it, leaving a projection of platinum wire at one end and copper wire at the other, the former only being brought in contact with the nutrient fluid. Other forms of electrodes will be mentioned later. The leads from the electrodes were connected with a standard condenser of one micro-farad and the condenser discharged through a galvanometer by means of a Morse key. This arrangement was adopted, on the suggestion of Mr. H. Morris Airey, to eliminate the resistance of the circuit. A Clark's cell discharged in the same manner through the galvanometer served to show the number of deflections upon the galvanometric scale, which corresponded to the standard voltage.

The electrodes were carefully tested, and it was quite certainly ascertained that no E.M.F. was developed at the junction of the platinum and copper. The cell when charged was also thoroughly tested to determine whether any E.M.F. produced could be due to differences in temperature between the media separated by the porous cylinder, to osmotic effects, or to evaporation. It might be supposed that a possible difference of temperature between the junctions of copper and platinum might give rise to a thermo-electric effect, but this point was decided by heating the outer fluid several degrees above that of the inner, and under these conditions no E.M.F. was developed. Again, many trials were made, by varying the concentration of the solutions outside and inside the porous cylinder, to prove whether the E.M.F. could be produced by reason of osmotic effect, but this was found not to be the case. Evaporation from either the inner or outer fluids was also effectually prevented, and thus any E.M.F. which might be developed could not be attributed to evaporation currents.

As oxygen is not liberated in any of the reactions considered in this investigation, no electric effects could be produced by oxidation of the platinum electrodes.

Platinum, it is well known, often contains a local charge, and a difference of potential might thus be registered upon the introduction of the two electrodes into the solutions. Experience showed that this local charge occurred, but from repeated tests with control cells it was proved to be constant during the limits of an experiment, and allowance for this effect could be made. This relative charge upon the platinum wire, however, is altered by friction, and it is essential to prevent any shaking or other disturbance of the electrodes during an experiment. The leads from the electrodes were therefore fixed to a support, so that no disturbance of the electrode was possible

when making the connections, and the difference of potential to be described could in no degree be attributed to friction inside the cell.

In every case the cells when set up were allowed to settle down and were tested before the commencement of any experiment. After these precautions against experimental error, the way seemed clear for a study of the actual effect produced by the vital activity of living organisms.

The fermentative activity of yeast upon solutions of sugar seemed to offer a promising subject for investigation, and ordinary commercial yeast in a very fresh condition, with glucose as a medium, was employed, because of the rapid and marked fermentation which is developed, and which one might expect to be reflected in a pronounced electrical effect.

For the study of the fermentation of yeast a very useful form of diaphragm can be constructed from parchment dialysing tubes, cut into convenient lengths. A glass cup tied firmly at one extremity and a glass ring at the other served to keep it distended, and as the tube could be discarded after each experiment, this kind of porous cylinder proved very convenient and was used in preference to a porous pot. The relative size of the cylinder and glass jar was such that when each contained 100 c.c. the level of the outer and inner fluids was approximately the same.

As an arbitrary standard it was found convenient to take 50 grm. of yeast and mix with 100 c.c. of water and to apportion 10 c.c. of this for each cell. By this method approximately the same quantity of yeast would be used for each cell, and the results of experiments would be comparable one with another.

The cell being set up, 100 c.c. of a given solution of glucose was poured into the cylinder and into the jar, the electrodes were inserted and the necessary connections made, and readings taken on the galvanometer determined the constant relative charge on the electrodes.

The yeast could now be introduced into the outer fluid, and after this was done readings of the galvanometer were taken at frequent intervals. For a short time after the introduction of the yeast no E.M.F. could be observed, but as the organism commenced its activity and the fermentation was set up, a gradually increasing voltage was registered, the rise of the voltage being determined by the concentration of the glucose solution, the temperature, and the quantity of yeast added.

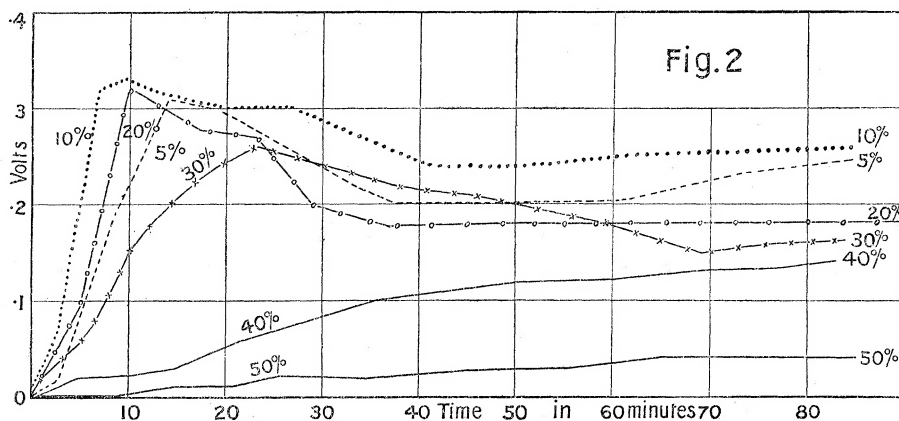
On disconnecting the yeast-glucose cell, and substituting for it a standard cell, it was always found from the swing of the galvanometer that the glucose solution with yeast acted as the zinc of an ordinary galvanic cell, the current passing in the cell from the yeast-glucose solution to the glucose solution. Moreover, when the inner fluid was inoculated instead of the outer fluid, the

seat of the chemical action became changed and the direction of the current was always reversed.

We may now proceed with some typical examples of the results obtained with experiments undertaken first with yeast.

Various Concentrations of the Medium.

With the ordinary standard of 5 gramm. of yeast at a temperature of 25°C . and a 10-per-cent. solution of glucose an E.M.F. of 0.32 volt was registered in the course of 7 minutes, after which the voltage gradually declined until at the end of 40 minutes it was 0.25, at which figure it remained fairly constant for a further period of 50 minutes. A reference to fig. 2 shows the variations of the voltage with solutions of different degrees of concentration.

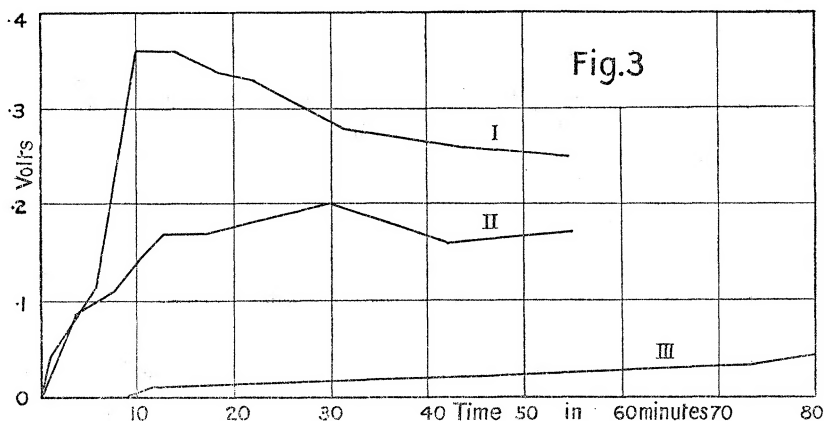


Curves showing development of E.M.F. in 5, 10, 20, 30, 40, and 50 per cent. solutions of glucose. 5 gramm. of yeast employed in each case. Initial temperature 25°C .

With 20-per-cent. glucose the maximum voltage, 0.32, was reached in 10 minutes; with 5-per-cent., 15 minutes was occupied in reaching a maximum of 0.31. The 10-per-cent. thus appeared to be the best mean, but it will be seen that the concentrations of 5, 10, and 20 per cent., though varying slightly in their initial rapidity of action, attained a very similar voltage, and the curve thereafter followed much the same course. With 30-per-cent. glucose there was a slower development, the maximum voltage, which was 0.26, being attained only after 23 minutes, while with still higher degrees of concentration a markedly slower action and lower voltage indicated a solution much less favourable to the development of the yeast. Thus 40-per-cent. glucose registered only 0.18 volt after 90 minutes, and 50-per-cent. 0.08 after the same period.

Varying Amounts of Yeast.

Fig. 3 gives the results of experiments to determine the effect of using different amounts of yeast. The three cells here given as an example were charged at the same temperature (23.5°C.) from the same solution of glucose, and to the first was added 5 gm. of yeast, to the second 2.5 gm., and to the third 0.5 gm. With 5 gm. of yeast the voltage rose rapidly to its maximum of 0.36 in 10 minutes. With 2.5 gm. the gradient of the curve



Curves showing development of E.M.F. with different amounts of yeast, in a uniform 10-per-cent. solution of glucose. I, 5 gm. of yeast; II, 2.5 gm.; III, 0.5 gm. Initial temperature 23.5°C.

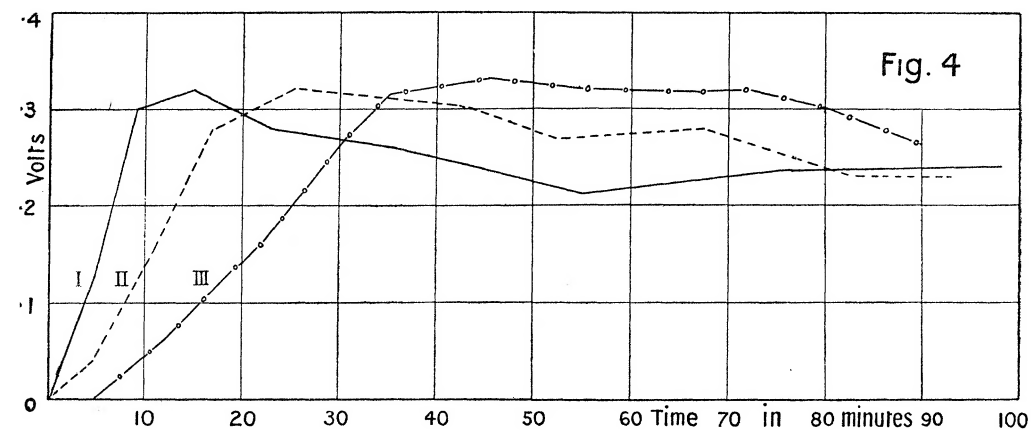
was much more gradual, reaching 0.3 volt at the end of 2 hours. With the minimum of 0.5 gm. the curve rose very slowly; at the end of 80 minutes it only showed 0.04 volt, and did not reach the maximum of 0.3 volt until after a period of 3 hours. A comparison of these curves gives striking evidence of the greater speed of reaction corresponding to the more active fermentation, *i.e.*, the greater number of active yeast-cells present.

Different Conditions of Temperature.

Under different conditions of temperature the results vary in a corresponding manner. This is well exemplified by the curves shown in fig. 4. When the yeast is introduced into a 10-per-cent. glucose solution at 25°C. a voltage of 0.3 is attained in 9 minutes, and the maximum, 0.32, in 15 minutes. But at 17°C. a voltage of 0.3 is not reached until after 20 minutes, and the maximum, 0.32, after 25 minutes. At 10°C. the start was very slow, and there was a much more gradual approach to the maximum, 34 minutes being required for the development of 0.3 volt and 45 minutes for the maximum.

It is interesting to note that after the maximum voltage has been attained there is always a slight drop in the curve, which was not so pronounced following the more gradual rise at the lower temperature.

When cells were maintained at a temperature of 50° C. there was no deflection of the galvanometer, and when the temperature was lowered to 25° C. there was again no electric effect, showing that the vitality of the yeast had been destroyed. At 0° C. no deflection could be detected, but when the temperature of these cells was gradually raised by placing the cell in an incubator, a voltage was soon observed which continued to increase as the temperature became more favourable. At the freezing point therefore the yeast was not killed, and the absence of electrical action at this temperature shows an effect of suspended animation.



Curves showing E.M.F. developed at different temperatures. 5 grm. of yeast added in each case to a 10-per-cent. solution of glucose. I, initial temperature 25° C.; II, initial temperature 17° C.; III, initial temperature 10° C.

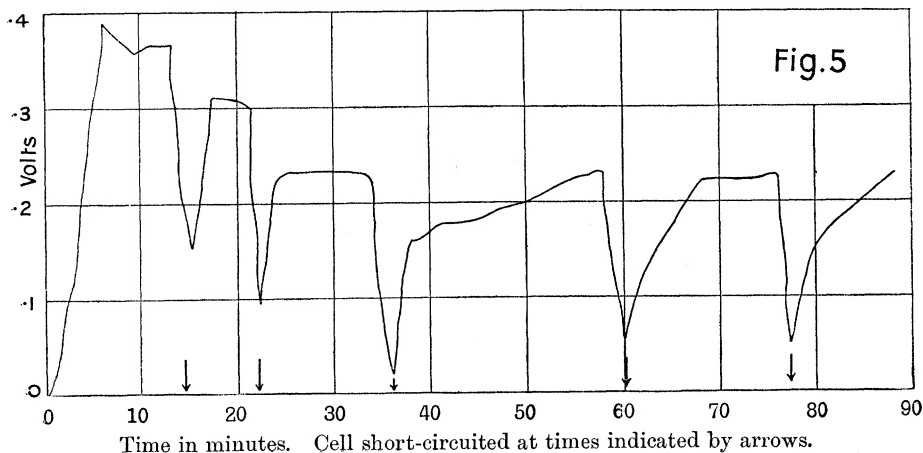
It should be mentioned that the initial temperature of the fermenting solution does not remain constant, but gradually changes as the process of fermentation proceeds.

These results demonstrate that the E.M.F. developed can only be attributed to the fermentative action of the yeast, and the electrical effects are the measure of the activity of the yeast. It has been shown that the character of the culture medium modifies to a great extent the electrical effects produced, and degrees of concentration which are too high for the favourable growth of the yeast are reflected in the diminution of the voltage registered and a much reduced speed of reaction. Further, the speed of reaction varies with the amount of active yeast present in the cells, and the gradient of the curve is directly influenced by this factor, being much steeper

when the organism is employed in a more effective strength. It was also found that the gradient of the curve indicating the growth of potential is very steep when the yeast is introduced into a solution of glucose at the optimum temperature, but the curve is much less steep when the temperature approaches the limits of the maximum or minimum for the organism. At the minimum temperature, 0°C. , and at the maximum, 50°C. , no E.M.F. is produced, and it is important to note that a difference of potential is only found within the limits at which the various organisms can live.

The maximum voltage recorded with yeast and glucose or cane sugar was 0.3—0.4, and a voltage of this order was never exceeded in any of the experiments under any of the conditions tried. When battery cells of a larger capacity, containing $1\frac{1}{2}$ to 2 litres, were used, it was found that no difference could be detected in the amount of the E.M.F. registered, and the voltage thus bears no relation to the volume of the fermenting liquor. It appears also that the voltage is quite independent of the thickness of the platinum wire or the surface of the electrode, thus when pieces of platinum foil up to $2\frac{1}{2}$ cm. square were welded on to the platinum wire, the voltage remained the same.

A small current can always be detected in the yeast-glucose cells, and the voltage as registered by metallic conductors is due to the charge collected in the fermenting liquid. When a short circuit is made by joining the leads, the liquid in the neighbourhood of the electrodes is at once discharged, and the voltage is considerably reduced. But when the leads are again separated the E.M.F. gradually increases as the liquid recovers its charge through the activity of the yeast cells, and it becomes once more fully developed in a time varying according to the species of organism and the cultural conditions. Fig. 5 shows the effect of a short circuit in diminishing the E.M.F.



and the recovery of the potential difference after separating the leads. 5 grm. of yeast were used, and a 10-per-cent. solution of glucose at 28° C., the short circuits being made at intervals of 15, 22, 36, 60, and 78 minutes after the introduction of the yeast.

Enzymes.

The important part which enzymes play in the activity of yeast during the breaking down of sugar into alcohol and CO₂ naturally leads one to an inquiry as to any electrical effects which may accompany their action, and to look for any such effects accompanying hydrolysis. At present I have had little opportunity for extended observations on this interesting question, but it may be worth while to record the results so far obtained. The two enzymes—invertase and diastase—with which some work has been undertaken, both exhibit distinct electrical response, though in a much smaller degree than has been already described.

Invertase.—For the study of invertase, a quantity of yeast was ground in a mortar with sand and lixiviated with a small quantity of water. After filtration through paper the filtrate was passed through porcelain to ensure that all yeast-cells were removed, and half of the solution was boiled to serve as a control.

Cells were charged with 5- and 10-per-cent. cane sugar, and the invertase solution added. In both cases a small voltage was registered, 0.02 for the 5-per-cent. and 0.03 for the 10-per-cent. The development of the E.M.F. was very gradual, and it was only after 70 minutes that the above readings were recorded. Treatment with Fehling's solution showed that inversion had taken place. No E.M.F. could be detected with the control cells to which the boiled solution was added, and tests with Fehling showed that the invertase had been destroyed.

Diastase.—A similar cell charged with a 0.5-per-cent. starch emulsion, to which was added 10 c.c. of a 5-per-cent. solution of diastase, gave an E.M.F. of 0.05 volt. The gradient of the curve of the potential development was again very gradual, a voltage of 0.3 being reached in ten minutes, after which it slowly increased.

Thus, in the hydrolysis of the polysaccharose starch to the monosaccharose glucose and fructose, a larger E.M.F. is produced than in the hydrolysis of the disaccharose cane sugar to the same monosaccharose.

Hydrolysis of Cane Sugar by means of Sulphuric Acid.

For this experiment a cell was charged with a 1-per-cent. solution of sulphuric acid. It was found that if water were added to either the inner

or outer fluid an E.M.F. might be developed, due to the variation of the concentration of the sulphuric acid on either side of the diaphragm, and to avoid this effect the cane sugar was added in the crystalline form. Following the addition of the sugar an E.M.F. was produced amounting only to 0.02 volt, the sugar and sulphuric acid being zincative. Testing with Fehling's solution showed that hydrolysis had taken place.

The small voltage developed when cane sugar is hydrolysed by dilute H_2SO_4 was further confirmed by adding the sugar as a thick syrup. In this case a battery of five cells in series was arranged, the temperature of the cells being 26°C . The syrup was added as quickly as possible to the cells, and readings taken at once. From this battery an E.M.F. of 0.1 volt was developed almost immediately, and too quickly to construct a curve showing the times of development of the E.M.F. This method, however, is useful in showing how the hydrolysis proceeds and the speed with which this reaction dies away.

Bacteria.

I may now briefly refer to some experiments which were made with certain species of bacteria. Pure cultures of *Bacillus coli communis*, *B. fluorescens*, *B. violaceus*, and *Sarcina lutea* were obtained from Dr. Kral, and, after sterilisation as previously described, inoculations were made from each of these micro-organisms upon nutrient solutions in the boiling tubes. The solution used was a modification of the well-known Pasteur's solution, viz. :—

Potassium phosphate	2
Calcium phosphate	0.2
Magnesium sulphate	0.2
Ammonium tartrate.....	10
Asparagine	0.5
Water	1000

B. coli communis flourished readily in this medium and proved to be an excellent subject for the investigation. At 30°C . this organism, when grown in the above solution, developed an E.M.F. recorded by the galvanometer as 0.308 volt. Using the same solution, but replacing the asparagine with 0.2-per-cent. starch, it gave rise to a voltage of 0.349. At 20°C . the voltage recorded was 0.534.

In the case of *B. violaceus*, *B. fluorescens*, and *Sarcina lutea* no E.M.F. was produced, these organisms causing no deflections of the galvanometer, either at 30°C . or at 20°C . It was found that the medium employed was unsuitable for the growth of these bacteria, and they had quite failed to develop under these conditions.

At this stage the action of yeast was also studied, and this proving a very convenient substance for experiment, further investigation of bacteria was for the present abandoned.

The employment of this form of galvanic cell may be usefully extended to the observation of other physiological processes, and it will be of interest to note some electrical effects indicated during the conversion of venous to arterial blood and *vice versa*.

A quantity of fresh warm blood was obtained, a part of which was poured into a parchment tube and part placed in the jar surrounding it. With a platinum electrode in the jar and in the parchment tube, it was found that arterial blood was zincative to venous blood. Many trials made, by alternately forcing air or carbonic acid through the blood in the jar or in the tube, showed that the oxygenating stream raised the potential of the blood, so that an electric current passed in the cell from the arterial to the venous blood; with the entering stream of CO_2 , however, the potential was lowered and an electric current was produced in the opposite direction. Thus the arterial and venous bloods possess a contrary electrical sign. As a corollary it would seem that this change of sign may play an important part in the electro cardiogram of the heart, and that when arterial is converted into venous blood this change of sign may be looked for as one of the accompanying events.

Comparison with a Galvanic Cell.

The study of the electrical effects during well-known chemical actions by means of a galvanic cell constructed on the principle already described is of much interest for comparison with the electrical effects of fermentation.

A glass jar containing a porous pot, into each of which a dilute solution of sulphuric acid is poured so that the level of the outer and inner fluids should be the same, a pair of platinum electrodes inserted one in the jar and one in the porous pot, and a little zinc added to either the outer or the inner fluid, constituted a cell comparable with the glucose-yeast cell previously described.

With such a cell, using a 10-per-cent. solution of sulphuric acid and 5 grammes of granulated zinc, the condenser and galvanometer gave an E.M.F. of 0.7 volt (see fig. 6). The direction of the current in the cell was from the sulphuric acid and zinc to the sulphuric acid, and thus followed the same course as that observed in the case of glucose and yeast. The curve indicating the E.M.F. in volts is also developed in a manner parallel to that of the yeast and glucose. The galvanometer showed no immediate effect following the introduction of the zinc, but during an interval of 8 minutes

the maximum voltage was gradually developed. This voltage died away by degrees as the zinc was dissolved and the chemical action ceased.

It should be noted that the zinc was not allowed to come in contact with the platinum electrodes, and thus the bubbles of hydrogen escaped from the zinc and not from the platinum wire.

A similar cell, in which hydrochloric acid was used in conjunction with zinc placed in the jar, gave parallel results, the voltage being 0.7.

Other substances used to develop an E.M.F. were phosphorus, potassium, and sodium.

Phosphorus.—The cell and porous pot were charged with distilled water, and a pair of platinum electrodes, one immersed in the outer and the other in the inner fluid, were connected with the condenser and galvanometer. The relative charge upon this pair of electrodes being determined, a stick of phosphorus was suspended partially immersed in the outer fluid, the temperature of the water in the jar being 10° C. For a period of 28 minutes no effect could be observed on the galvanometer, the constant relative charge upon the pair of electrodes giving the same number of deflections, although copious fumes were being given off. At the end of this period the effect of the oxidation could be observed, and the voltage developed steadily increased. After 11 minutes it increased to 0.05, after 13 minutes to 0.06, and after 35 minutes to 0.1. After this time, the phosphorus, becoming dry, ignited, and the E.M.F. then suddenly increased to 0.21 volt. It should be noted that the current was in the same direction as with zinc and sulphuric acid, namely, the oxidising phosphorus was zincative.

In this experiment the comparatively long period between the introduction of the phosphorus and the first signs of the development of any E.M.F. was due to the low temperature of the cell (10° C.), and with a higher temperature it was found to be considerably reduced.

Potassium and Sodium.—A similar cell, with water in both jar and porous pot and with a pair of platinum electrodes, gave an E.M.F. when either metallic potassium or sodium was introduced into the jar, the direction of the current being the same as in the other cases already mentioned.

From the curves given it will be noted that a short interval of time elapses after the introduction of the zinc into the sulphuric acid before the E.M.F. can be detected, although the action of the acid upon the zinc is at once evidenced by the liberation of the hydrogen or the fumes given off from the phosphorus. A similar time of presentation was observed with both sodium and potassium. Although experiments have not been undertaken with special regard to this point, it is suggested that the gradient of the curve of the E.M.F. is a function of the temperature.

Thus, a study of this cell when charged with dilute sulphuric acid and zinc, or with water and either phosphorus or potassium or sodium, gains importance from its bearing upon the action of yeast and glucose in a similar cell. It has been proved that the principle of this special type of cell is equally applicable to the two cases, and the results obtained are precisely analogous in both sets of experiments. For the purposes of the present research it is sufficient to have established this point, to note that an E.M.F. is developed in these special cells, and that the centre of chemical activity with platinum electrodes is always zincative.

Non-polarisable Electrodes.

The behaviour of non-polarisable electrodes in a cell of this construction is also of much interest, and an exact parallel is found between the action of these electrodes when used in conjunction with either a yeast-glucose or a zinc-sulphuric acid cell.

The non-polarisable electrodes consisted of a glass U-tube, plugged at the bend with kaolin; into one limb of this tube a saturated solution of zinc sulphate was poured and a rod of amalgamated zinc inserted, the zinc being connected with the condenser and galvanometer leads. Into the other limb was placed a solution of glucose or of sulphuric acid of the same concentration as that employed in the cell, and a lamp wick soaked in the glucose or sulphuric acid solution was arranged so that one end of it dipped into the U-tube and the other into the cell.

Another form of non-polarisable electrode consisted of a pair of glass tubes, closed at one end either with a plug of kaolin or with a piece of bladder tied tightly round. These tubes were partially filled with the zinc sulphate solution, into which dipped a rod of amalgamated zinc. One of the tubes was inserted into the outer and the other into the inner fluid of the cell, and the connections were made with the condenser-galvanometer circuit as before.

Such electrodes, when used either with a yeast-glucose cell or a zinc-sulphuric acid cell and the condenser, gave no deflections upon the galvanometric scale. In both these cases, however, the existence of an electric current, depending upon the resistance of the circuit, can be shown by connecting the leads from the non-polarisable electrodes directly with the galvanometer. The electrical action is developed both in the yeast-glucose and in the zinc-sulphuric acid cells, as shown by the current generated, but the condenser method is not applicable with the use of non-polarisable electrodes.

As a further test of this principle, cells were set up with a pair of

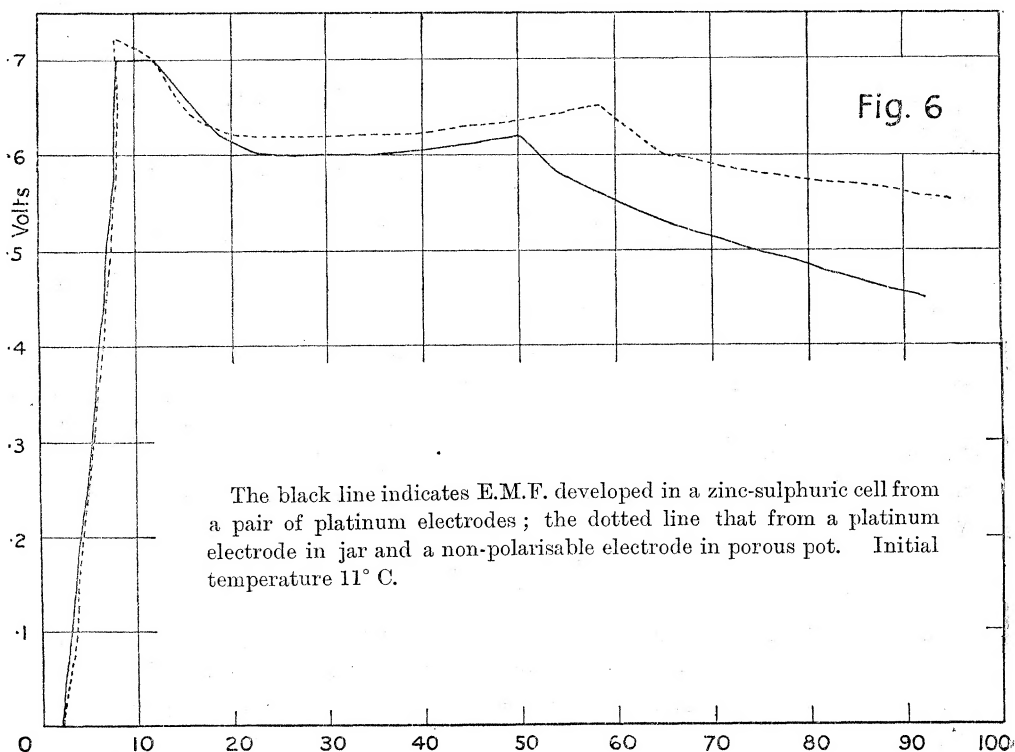
platinum electrodes and a pair of non-polarisable electrodes. With this arrangement four possible combinations of the electrodes are possible:—

- I. A pair of platinum electrodes.
- II. A pair of non-polarisable electrodes.
- III. A platinum electrode in the jar and a non-polarisable one in the porous pot.
- IV. A non-polarisable electrode in the jar and a platinum one in the porous pot.

With a zinc-sulphuric acid cell, in which the constancy of the relative charges upon the four pairs of electrodes had been determined, the following facts were observed when the zinc was added to the sulphuric acid in the jar:—

I. Platinum-platinum electrodes. After a short interval an E.M.F. was developed which soon attained its maximum, and then gradually died away as the zinc was consumed (fig. 6).

II. Non-polarisable electrodes in both jar and porous pot. No E.M.F. was developed; that is, the discharge of the condenser through the galvanometer



produced no effect. When, however, these electrodes were connected directly with the galvanometer the spot immediately moved "off scale," showing the existence of an electric current developed in the cell, the strength of which depended upon the resistance of the circuit.

III. Platinum in the jar, non-polarisable electrode in the porous pot. An E.M.F. was developed of much the same voltage as in I, as shown by a comparison of the curves in fig. 6.

IV. Non-polarisable electrode in the jar and platinum in the porous pot. No E.M.F. could be detected with the condenser, but an electric current was evident as in II.

It should be remarked that the relative charge upon the electrodes in the cases III and IV was, relatively speaking, large; thus in III the number of deflections showed a difference of potential of 1 volt, and also in IV a difference of potential of 1.1 volt. The deflections on the scale of the galvanometer were of opposite sign, that in III being anti-zincative, and that in IV zincative. In III the voltage due to the relative charge decreased as the zinc was acted upon by the sulphuric acid, at first very rapidly, until the maximum E.M.F. was produced by the zinc and sulphuric acid. Following this it increased gradually as the action ceased; and after some 12 hours, when all the zinc had been dissolved, the relative charge showed only a slight difference from its former voltage. In this case the E.M.F. produced by the solution of the zinc was opposite in sign to the relative charge of the pair of electrodes.

In IV the relative charge remained constant throughout the duration of the experiment.

A cell charged with yeast and glucose and a similar four pairs of electrodes gave results exactly parallel with those described for zinc and sulphuric acid:—

I. Platinum electrodes. The curve of the E.M.F. with galvanometer and condenser developed in the normal manner.

II. Non-polarisable electrodes. No deflections of the galvanometer were to be observed when the condenser was discharged through it, but a very distinct current was registered when the electrodes were connected directly with the galvanometer.

III. This gave the normal curve of the E.M.F. as in I.

IV. The same effects as in II were indicated.

All these tests combine to show quite clearly that, though a current is certainly generated, no voltage can be registered with non-polarisable electrodes, and this special form of electrode is unsuitable for use with the condenser.

Other Electrodes.—Although platinum wires were employed as the standard electrodes throughout this investigation, trials were also made with other metallic wires such as gold, nickel, tin, zinc, and aluminium. All these metals when used as electrodes, as well as carbon plates from Leclanché cells, showed the existence of a difference of potential between the yeast-glucose solution and the glucose solution, and also the existence of an electric current passing in the cell from the yeast-glucose to the glucose. The amount of the E.M.F., however, was found to vary with the different electrodes used; but an inquiry into the causes of this variation leads into the domain of physical chemistry and lies outside the scope of the present investigation.

It may be mentioned that a battery of six cells with carbon electrodes connected in parallel gave a current of 1.25 milliampère.

In this preliminary investigation only commercial yeast of the kind known as "German yeast" was used, and at present no experiments have been made with pure cultures of the different races of *Saccharomyces*. So far cane sugar and ordinary glucose dissolved in distilled water have provided the nutrient media for the yeast, but evidence is not wanting that very slight differences in chemical composition directly affect the character of the results obtained. Enough has been done to show that this electrical method may be conveniently utilised to study the vital activity of yeast and other organisms when growing under special cultural conditions. It will be a matter of interest to compare the E.M.F. produced by the different races of *Saccharomyces* in the various kinds of sugars, and also to study the effect of inorganic salts, such as phosphates, nitrates, &c. To follow up the question as it affects enzymes opens up a large field of work, and this subject presents a problem in itself. The action of bacteria in producing electrical effects also awaits more detailed investigation.

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Summary and Conclusions.

The disintegration of organic compounds by micro-organisms is accompanied by the liberation of electrical energy.

The difference of potential was investigated by the employment of a special type of galvanic cell with platinum electrodes, by means of which the electrical charges set free in the vital processes of the micro-organisms were

collected and transferred to a condenser. Since CO_2 is the only gas liberated in alcoholic fermentation there is assumed to be no oxidation of the platinum electrodes. The charge, as measured by a ballistic galvanometer, was found to correspond to an E.M.F. of 0.3 to 0.5 volt between the fermenting and non-fermenting fluids. A difference of potential also occurs during hydrolysis, either by enzymes or by weak acids.

Various metallic electrodes can be used in conjunction with a condenser for the estimation of the E.M.F. produced. Non-polarisable electrodes, however, give no voltage with a condenser and are unsuitable for this method. Distinct electric currents are found whichever form of electrode is used.

The electrical effects are an expression of the activity of the micro-organisms and are influenced by temperature, concentration of the nutrient medium, and the number of active organisms present. These effects are only found within the limits of temperature suitable to the micro-organisms and under conditions which are favourable to protoplasmic activity.

The maximum voltage recorded was 0.3 to 0.5 volt, and a voltage of this order was never exceeded in any of the experiments undertaken with micro-organisms.

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